Fungal Genetics Reports

Volume 14 Article 15

Glycerol as carbon source in induction studies

W. K. Bates

Glycerol as carbon source in induction studies

Abstract
Glycerol as carbon source in induction studies

Bates, W. K. Glycerol as carbon source for induction studier in Neurospora.

In studies of enzymes induced by lactose and by galactose in Neurospora, it has become apparent that growth conditions must be carefully controlled, and that shaking cultures containing a single carbon source con provide reproducible conditions well-suited for there studier. The ideal carbon source should cause no repressive interference with the induction process. Glycerol is suitable for such studies, but wild type strains ore quite variable in their ability to grow on glycerol under the required conditions. The isolate 105-L5-A (formerly designated L5D, Bates ond Woodward 1967 Neurospora Newsl. 12: 11) shows greatly improved growth on glycerol when compared with STA4. Crosser of thir isolate to wild type 74-OR8-1a (Bates 1967 Genetics 56: 543) yielded a variety of isolates with improved glycerol growth characteristics, although initial selection was for lactose growth. Two of there were crossed (211-L5-a x 341-&A) ond an isolate designated 41 I-L5-A was obtained. This isolate has been used for all subsequent glycerol growth studier.

Growth conditions ore: rotary shaking, 3/4 inch radius, 150 cycles per minute, 30 ± 0.5°C, 0. 18 M glycerol, Vogel's medium, 200 ml in 500 ml Erlenmeyer flasks, mounted at a 30 degree angle. The Vogel's medium is autoclaved at 2 x concentration, and the carbon source is autoclaved separately in 100 ml water. The inoculum is 106 conidio per ml medium. Under there conditions growth is linear for ca. 90 hours (yielding co. 1.5 g dry weight), ond comparisons are mode by harvesting at 48 hours (yielding co. 0.7 g dry weight). The rote of growth with glucose under there conditions is ca. two timer the rate obtained with glycerol.

In comparison with 411-L5-A, token as 100%, growth of some wild type strains on glycerol can be grouped in the following way: STA4 and RL-A, 33-36%; ST73 a, RL-a ond Em-A (FGSC#691), 58-68%; Em-o (FGSC#692), 103% (all based upon total mycelial dry weight at 48 hrs). RL-A and RL-a are Rockefeller-Lindegren isolates obtained from J. F. Wilson. It is apparent that a mating type shows better glycerol growth than doer A for all three strains, except for 411-L5-A. When grown on sucrose under otherwise identical conditions, little difference (± 10%) in total growth is observed among there different strains. Another distinguishing characteristic is the orange pigmentation which occurs under these glycerol growth conditions in on inverse relationrhip to ability to grow on glycerol. The Em-o and 411-L5-A cultures show no evidence of this pigmentation.

Among the isolates obtained along with 41 I-L5-A, there was a marked correlation between ability to grow on glycerol and reduced production of conidia. For example, 411-L5-A producer only 30-50% of the conidia produced bySTA4 when grown ond harvested under the same conditions. That this characteristic is not necessarily associated with glycerol growth is shown by Em-o, which conidiates more abundantly than STA4, but which grows well on glycerol. Crosses designed to combine the glycerol growth characteristics with amino acid and inositol requirements are now in progress. The 411-L5-A isolate producer abundant protoperithecia on Westergaard's synthetic cross medium, and up to 90% spore viability, but the mature perithecio apparently have low internal pressure, and discharge ascospores weakly.

It appears that the 411-L5-A amino acid auxotrophs hove very similar glycerol growth characteristics, but there studier hove not been completed. Such characteristics would allow very precisely controlled studier of incorporation of labeled amino acids during induction studier. If there isolates appear to be potentially useful to other workers, the set of cultures will be deposited in the Fungal Genetics Stock Center. (Supported by NSF Grant GB 5189). - - - Department of Biology, The Univ. of North Carolina at Greensboro, Greensboro, North Carolina 27412.